



Development of RT-semi-nested PCR for detection of hepatitis A virus in stool in epidemic conditions.

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Titre	Development of RT-semi-nested PCR for detection of hepatitis A virus in stool in epidemic conditions.
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Auteur	Apair-Marchais, Véronique [1], Ferre, Virginie [2], Colonna, F. [3], Dubois, F. [4], Ponge, A. [5], Billaudel, S. [6]
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Résumé en anglais	<p>The purpose of this study was to determine the efficiency of semi-nested PCR in detecting hepatitis A virus (HAV) RNA. During a 2-year period (1990-1991), HAV RNA was searched for in shellfish from the French Brittany coasts using cRNA and vRNA probes. In January 1992, at the time of a hepatitis A outbreak, 28 stool samples were collected from infected patients (18 adults, 10 children) with anti-HAV IgM. Four samples from subjects with negative HAV serology were used as negative controls. Nucleic acid amplification (reverse-transcription-semi-nested PCR) was performed to detect HAV in stool. HAV RNA was purified by phenol-chloroform extraction and converted to cDNA using reverse transcriptase (Mu-MLV). After amplification, PCR products were visualized on an ethidium-bromide-stained gel and confirmed by hybridization with a specific digoxigenin-labelled oligoprobe. Samples were also studied by molecular hybridization with cRNA and vRNA probes. After onset of the illness, HAV RNA was detected over a longer time period by semi-nested PCR (16/28) than by hybridization (0/28). Even though biological diagnosis of hepatitis A will continue to rely on the detection of anti-HAV IgM, PCR should be useful in certain clinical cases (diagnosis of relapse) and for epidemiological and environmental monitoring of viruses.</p>
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